

**ART 34 AMDT**CLAIMS

1. An isolated polynucleotide comprising a nucleotide sequence expressed at the mRNA level in human mononuclear leukocytes having cell-surface TNF receptor, thereby increasing cleavage and release of the receptor from the surface of the cell.
2. The polynucleotide of claim 1, wherein the nucleotide sequence is contained in a sequence selected from the group consisting of
  - a) SEQ. ID NO:1;
  - b) SEQ. ID NO:2 and SEQ. ID NO:3;
  - c) SEQ. ID NO:4;
  - d) SEQ. ID NO:5;
  - e) SEQ. ID NO:6;
  - f) SEQ. ID NO:7;
  - g) SEQ. ID NO:8;
  - h) SEQ. ID NO:9; and
  - i) SEQ. ID NO:10.
3. An isolated polynucleotide comprising at least 30 consecutive nucleotides in said nucleotide sequence of a polynucleotide according to any of claims 1-2.
4. An isolated polynucleotide comprising a linear sequence of at least 50 consecutive nucleotides at least 90% identical to a sequence contained in said nucleotide sequence of the polynucleotide of claim 1.
5. An isolated polynucleotide of at least 50 nucleotides capable of hybridizing specifically to said nucleotide sequence of a polynucleotide according to any of claims 1-3 at 68°C in 0.5 M phosphate buffer pH 7, 7% SDS, and 100 µg/mL salmon sperm DNA, followed by washing in a buffer containing 3X SSC.
6. An antisense polynucleotide or ribozyme comprising at least 10 consecutive nucleotides in said nucleotide sequence of a polynucleotide according to claim 1 or 2, which inhibits the expression of a TRRE modulator.
7. An isolated polypeptide comprising an amino acid sequence encoded by a polynucleotide according to any of claims 1-5.

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8. The polypeptide of claim 7, selected from the group consisting of SEQ. ID NOS: 147-154.
9. An isolated polypeptide, comprising at least 10 consecutive residues in said amino acid sequence of a polypeptide according to claim 7 or 8.
10. An isolated polypeptide, comprising at least 15 consecutive amino acids which are at least 80% identical to a sequence contained in said amino acid sequence of the polypeptide according to claim 7 or 8.
11. The polypeptide of claim 7-10, which when incubated with COS-1 cells expressing TNF receptor, promotes enzymatic cleavage and release of the receptor.
12. The polypeptide of claims 7-11, which either:
  - a) lacks a membrane spanning sequence; or
  - b) is produced by a process comprising recombinant expression in a host cell followed by purification of the polypeptide from medium in which the cell is cultured.
13. A method of producing the polypeptide according to any of claims 7 to 11, comprising the steps of:
  - a) culturing host cells genetically altered to express the polynucleotide according to claim 3; and subsequently
  - b) purifying the polypeptide from the cells.
14. The method according to claim 13, comprising harvesting culture medium following step a); and purifying the polypeptide from the culture medium by a process comprising affinity chromatography.
15. An isolated polynucleotide encoding the polypeptide of claim 8 or 9.
16. An isolated antibody specific for a polypeptide according any of claims 7-11.
17. A method for producing the antibody according to claim 16, comprising immunizing a mammal or contacting an immunocompetent cell or particle with a polypeptide according to claim 9 or 10.

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18. An assay method of determining altered TRRE activity in a cell or tissue sample, comprising the steps of:
- contacting the sample with the polynucleotide of claim 4 or 5 under conditions that permit the polynucleotide to hybridize specifically with nucleic acid that encodes a modulator of TRRE activity, if present in the sample, and
  - determining polynucleotide that has hybridized as a result of step a), as a measure of altered TRRE activity in the sample.
19. An assay method for determining altered expression of a modulator of TRRE activity in a cell or tissue sample, comprising the steps of:
- contacting the sample with the antibody of claim 16 under conditions that permit the antibody to bind the modulator if present in the sample, thereby forming an antibody-antigen complex; and
  - determining complex formed in step a), as a measure of the modulator.
20. A method for assessing a disease condition associated with altered TRRE activity in a subject, comprising determining altered TRRE activity in the sample from the subject according to claim 18, or determining altered expression of a TRRE modulator according to claim 19, and then correlating the extent of alteration with the disease condition.
21. A method for decreasing signal transduction from a cytokine into a cell, comprising contacting the cell with a polypeptide according to any of claims 7-8 and 11-12, or with a polynucleotide according to any of claims 1-3 and 15.
22. A method for increasing signal transduction from a cytokine into a cell, comprising contacting the cell with a polynucleotide according to claim 6, or with an antibody according to claim 18.
23. The method according to claim 21 or claim 22, wherein the cytokine is TNF.
24. A method for screening polynucleotides for an ability to modulate TRRE activity, comprising the steps of:
- providing cells that express both TRRE and the TNF-receptor;
  - genetically altering the cells with the polynucleotides to be screened;
  - cloning the cells genetically altered in step b); and
  - identifying clones that enzymatically release the receptor at an altered rate.

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25. A method for screening substances for an ability to affect TRRE activity, comprising the steps of:
- incubating cells expressing TNF receptor with a polypeptide according to claim 9 in the presence of the substance;
  - incubating cells expressing TNF receptor with a polypeptide according to claim 9 in the absence of the substance;
  - measuring any TNF receptor released from the cells in steps a) and b); and
  - correlating an increase or decrease of the receptor released in step a) relative to that in step b) with an ability of the substance to enhance or diminish TRRE activity.
26. Use of a polypeptide according to any of claims 7-8 or 11-12, in the preparation of a medicament for treatment of the human or animal body by surgery or therapy.
27. Use of a polynucleotide according to any of claims 1-3, 6, or 15 in the preparation of a medicament for treatment of the human or animal body by surgery or therapy.
28. Use of an antibody according to claim 16, in the preparation of a medicament for treatment of the human or animal body by surgery or therapy.
29. Use of a polypeptide according to any of claims 7-8 and 11-12, a polynucleotide according to any of claims 1-3 and 15 or an antibody according to claim 16, in the preparation of a medicament for treatment of a disease selected from the group consisting of heart failure, cachexia, inflammation, endotoxic shock, arthritis, multiple sclerosis, and sepsis.
30. A method of treating cancer in a subject, comprising increasing signal transduction from TNF into cells at the site of the cancer in the subject according to claim 22 or 23.
31. A method of treating a disease selected from the group consisting of heart failure, cachexia, inflammation, endotoxic shock, arthritis, multiple sclerosis, and sepsis, comprising decreasing signal transduction from TNF into cells at the site of the disease in the subject according to claim 21 or 23.
32. The method of claim 31, comprising administering to the subject an effective amount of the polypeptide of any of claims 7-8 or 11-12.

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33. The polynucleotide according to any of claims 1-5, wherein said nucleotide sequence is not contained in any of the sequences of the following GenBank Accession Nos: AJ003355, AA806165; AI002979; T33896; U52522; AA779203; C06247; AA707194; AA599596; 5453538; U13369; and J03528.
34. The polypeptide according to any of claims 7-10, the sequence of which is not completely encoded by a polynucleotide sequence contained in any of the sequences of the following GenBank Accession Nos: AJ003355, AA806165; AI002979; T33896; U52522; AA779203; C06247; AA707194; AA599596; 5453538; U13369; and J03528.
35. The polynucleotide according to claim 15, the sequence of which is not contained in any of the sequences of the following GenBank Accession Nos: AJ003355, AA806165; AI002979; T33896; U52522; AA779203; C06247; AA707194; AA599596; 5453538; U13369; and J03528.
36. The isolated polynucleotide of claim 1, wherein the nucleotide sequence is expressed at the mRNA level in Jurkat T cells; and when COS-1 cells expressing TNF receptor are genetically altered to express the sequence, the cells have increased enzymatic activity for cleaving and releasing the receptor.

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